



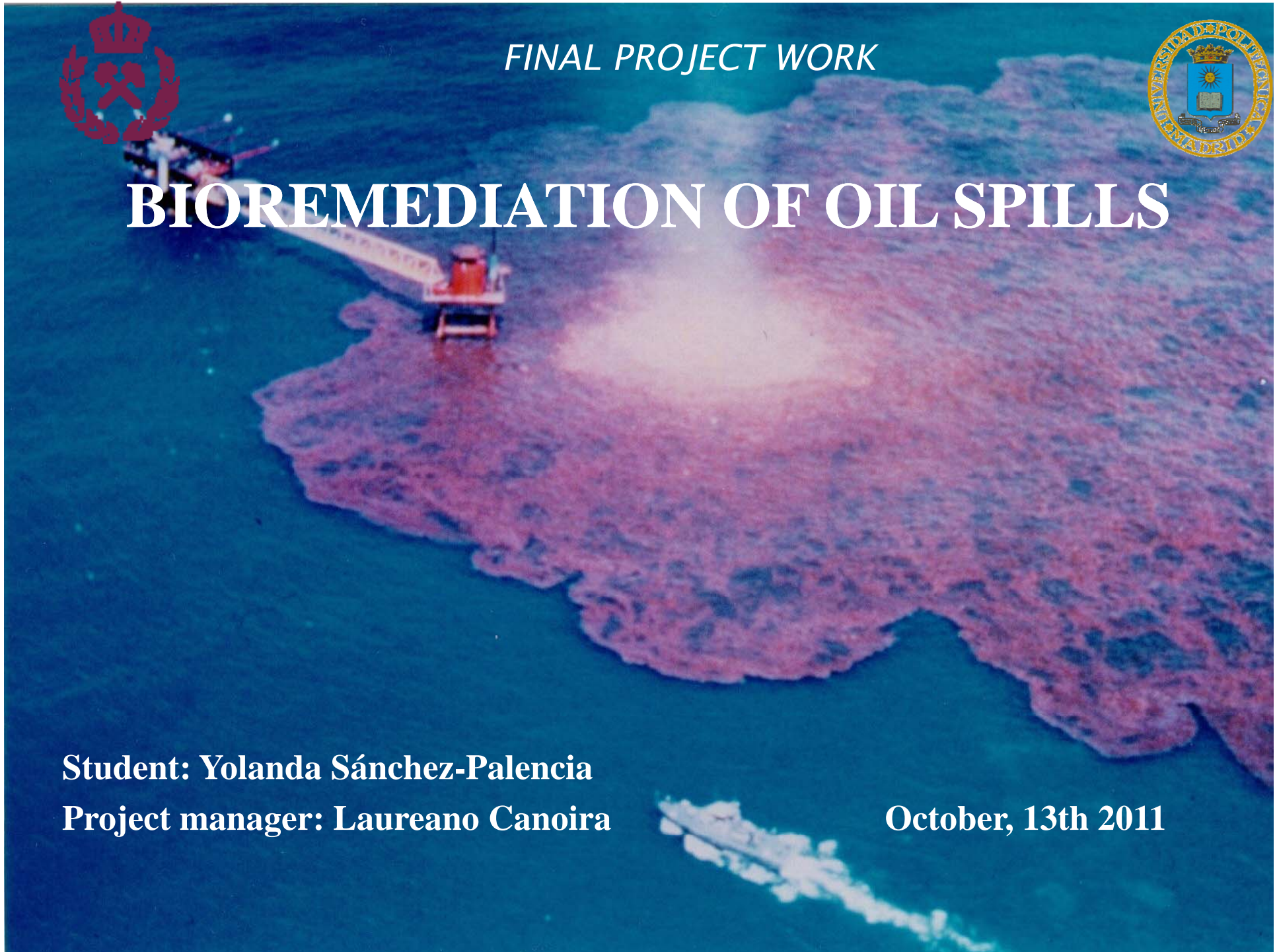
FINAL PROJECT WORK



BIOREMEDIATION OF OIL SPILLS

Student: Yolanda Sánchez-Palencia
Project manager: Laureano Canoira

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BACKGROUND

- The world demand for oil in 2008 was 85.62 million barrels per day (OPEC 2009).
- It was estimated that about 2 to 10 millions of tons of crude petroleum oil enter marine environments annually.
- Causes: Transport, tanker accidents, municipal and industrial wastes and runoffs.



PETROLEUM AND MICROBIAL DEGRADATION OF HIDROCARBONS IN MARINE ENVIRONMENTS

- *In natural ecosystems depends on the nature of the oil, on the composition of the indigenous microbial community, and on environmental factors (e.g., temperature, PH, nutrients...)*
- *Biodegradability of the oil components: n-alkanes>branched-chain alkanes>branched alkenes>lowmolecular- weight n-alkyl aromatics>monoaromatics>cyclic alkanes>polycyclic aromatic hydrocarbons (PAHs) >> asphaltenes*
- *The hydrophobic nature of PAHs makes their clean-up extremely difficult as they persist for a long period of time.*

PETROLEUM AND MICROBIAL DEGRADATION OF HIDROCARBONS IN MARINE ENVIRONMENTS

- *Examples of strains able to degrade petroleum:*
 - *benzene, toluene, ethylbenzene and xylene are Pseudomonas, Rhodococcus and Ralstonia*
 - *polyaromatic hydrocarbons such as:*
 - *naphthalene: Pseudomonas*
 - *phenanthrene : Pseudomonas and Haemophilus*
 - *anthracene : Rhodococcus*
 - *pyrene: Haemophilus and Mycobacterium*
 - *benzo[a]pyrene: Rhodococcus and Mycobacterium*

PETROLEUM AND MICROBIAL DEGRADATION OF HYDROCARBONS IN MARINE ENVIRONMENTS

- *characteristics that define hydrocarbonutilizing microorganisms:*
 - (1) *membrane-bound groupspecific oxygenases*
 - (2) *mechanisms for optimizing contact between them and the waterinsoluble hydrocarbons*
- *metabolize hidrocarbons to harmless end products such as CO₂ and water*
 - *Genera: Alcanivorax, Cycloclasticus, Oleispira, Oleiphilus, Thalassolituus...*

BIOAUGMENTATION AND BIOSTIMULATION

- **Bioaugmentation:** *addition of pre-adapted microbial cultures, single strains or consortia.*
 - *Consortia showed higher biodegradation rates since it provides the metabolic diversity needed*
- **Biostimulation:** *injection of nutrients (N, P...) and other supplementary components to the native microbial population.*
 - *Biosurfactants: solubilise hydrocarbon contaminants increasing availability for degradation.*

The decision to implement either or both of these techniques depends on the degrading capability of the indigenous microbes and the extent of contamination of the site.

BIOAUGMENTATION AND BIOSTIMULATION

➤ **Advantages:**

- *Economical and eco-friendly*
- *clean-up technique for heavy metals and/or organic pollutants*

➤ **Disadvantages:**

- *In real cases, the introduced population starts decreasing after being added due to stresses. (temperature, pH, nutrients, competition between introduced and indigenous biomass, etc.)*
- *higher concentrations of N and P sources in biostimulation can cause eutrophication, enhancing algal growth and reducing the oxygen concentration in the water*

PRODUCTION OF LIPIDS AND STORAGE COMPOUNDS BY HCB BACTERIA

- *Oil pollution is a temporary condition of carbon excess coupled to a limited availability of nitrogen that prompts marine oil-degrading bacteria to accumulate storage lipid compounds such as polyhydroxyalkanoates (PHAs), triacylglycerols (TAGs), or wax esters (WEs)*
- *production and export of lipids by Alcanivorax is a step in the synthesis of biosurfactants*

DISPERSANTS AND OIL MINERAL AGREGATES

Dispersants:

- *Is the most frequently employed method because can be applied to large oil spills and is more cost-effective than physical methods.*
- **Their property is to disperse oil into water enhancing the biodegradability due to the increased exposed surface of the spills to the agents**
- **Drawbacks:**
 - *many of the effective ones are toxic and/or not biodegradable*
 - **It is not a sustainable technique since it does not remove oil from the sea and just transfers oil on water layer**

DISPERSANTS AND OIL MINERAL AGGREGATES

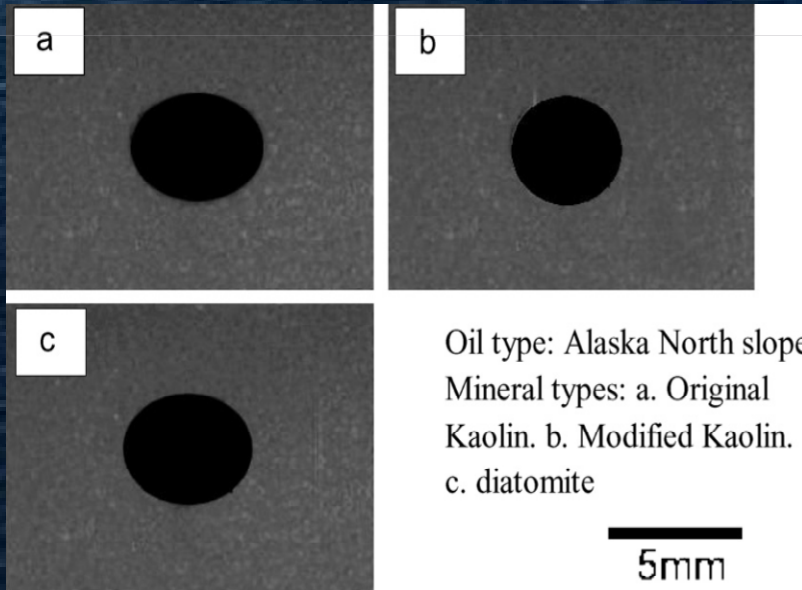
Oil-mineral aggregate (OMA):

- *Formation of OMAs is an effective alternative for oil spill cleanup*
- **The oil droplets trapped in OMAs were stabilized and prevented from re-coagulating, reducing the size and exhibiting higher biodegradation**
- *Oil associated with OMA has less chance to adhere onto bedrock and facilities*
- *the efficacy may be improved by dispersants*

DISPERSANTS AND OIL MINERAL AGGREGATES

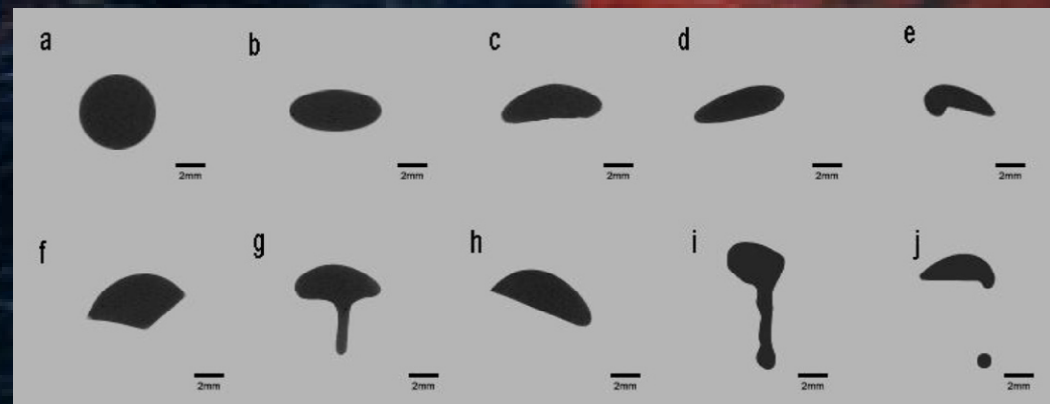
- *mechanisms of crude oil–mineral interactions:*
 - *hydrophilic minerals (Kaolin) have less oil–mineral interaction than the hydrophobic ones (calcite)*
 - *the polarity of the oils plays an important role due to negatively charged particles have an affinity to polar compounds of the oil*
- *The interaction between oils and minerals can be reflected by the shape and movement of oil droplets.*

DISPERSANTS AND OIL MINERAL AGGREGATES



a and c are hydrophilic minerals, b is a hydrophobic mineral

Effect of dispersants on the shape of an oil droplet



COLD ENVIRONMENTS

- *The same levels of contamination have a greater impact on these environments, as they have adapted to harsh conditions making them more sensitive*
- **Low ambient temperatures increase viscosity of oil, reduce evaporation of volatiles, and delay biodegradation**
- **Bioremediation normally takes place in the active layer on the top of permafrost; therefore, the effectiveness depends on some factors**

COLD ENVIRONMENTS

- *Factors affecting bioremediation*
 - **Temperature:** *Although the microbial biodegradation activity does not cease at subzero temperatures, the optimum is 15–30 °C for aerobic processes and 25–35 °C for anaerobic processes*
 - **Oxygen:** *Is a constraint in frozen ground because is scarce and his diffusion is partly or completely blocked becoming an anaerobic soil.*

COLD ENVIRONMENTS

- **Alternate electron acceptors:** *aerobic organisms utilize oxygen as electron acceptor. Anaerobes utilize nitrates, sulfates, CO₂, and ferrous metals*
- **Nutrients:** *nitrogen and phosphorus are in low concentration*
- **Toxicity:** *Lichens and mosses suffer heavy mortality. With higher air temperatures, more toxic components are lost through weathering*

COLD ENVIRONMENTS

➤ *Techniques*

- *Biostimulation: favorable effect of nutrient addition when aeration was provided*
- *Bioaugmentation: augmentation with cold-adapted biodegraders was unsuccessful*
- *snow, ice, and frozen ground can act as physical containments or barriers to limit the rate, and extent of oil spills*

An aerial photograph of a large ship, likely a research vessel, in the middle of the ocean. The ship is dark-colored with a white superstructure. A large, light-colored net or trawl is being deployed from the ship, creating a wide, circular pattern in the water. The water is a deep blue color. The text "THANK YOU FOR YOUR ATTENTION!" is overlaid in yellow, italicized font, with a small white square icon to the left of the first line.

 *THANK YOU FOR
YOUR ATTENTION!*